CXI. CAROTENE AND VITAMIN A.

THE TRANSFORMATION OF CAROTENE INTO VITAMIN A AS SHOWN BY A STUDY OF THE ABSORPTION SPECTRA OF RAT-LIVER OILS.

BY NORMAN STEWART CAPPER.

From the Sir Donald Currie Laboratories, Queen's University, Belfast.

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MOORE [1929, 1930] has advanced evidence to show that when highly purified carotene is administered to rats it is not stored in the liver in an unchanged condition, but suffers conversion to vitamin A. Among other evidence in favour of this view, preliminary results obtained by the present author in regard to the absorption spectra of the liver oils of rats receiving carotene were quoted. These preliminary experiments have now been confirmed and the results may be described in detail:

EXPERIMENTAL.

The liver oils, extracted by Dr Moore, were forwarded to me in sealed tubes. After weighing, the oils were dissolved in dry ether or chloroform and their absorption spectra determined with a Hilger E 3 all-quartz spectrograph used in conjunction with a Hilger sector photometer with a condensed copper spark as light source.

The numbering of the rats corresponds with that of Moore's paper [1930]. Closely similar absorption curves were obtained for the liver oils of rats receiving similar diets and representative examples are reproduced in Fig. 1.

The results are shown in Table I.

Table I.

No. and sex of rat	Diet	Solvent	Band at 325 $\mu\mu$	Band or inflexion in region of 280 $\mu\mu$
5 ♂	Vitamin A-free	Ether	No band	Inflexion
8 ♀	"	Chloroform	"	"
9 ♂	"	"	"	"
10 ♀	"	Ether	"	"
16 ♀	Vitamin A-free + 0.75 mg. per day carotene. M.P. 178°	Ether	Good band	Shallow band
17 ♀		Chloroform	"	"
19 ♂	Vitamin A-free + red palm oil	Chloroform	"	Inflexion
20 ♀	(1.5 g.)	Ether	"	Shallow band
22 ♂	Vitamin A-free + fresh carrots <i>ad lib.</i>	Chloroform	"	Shallow band

DISCUSSION.

It is seen that in every case the liver oils of rats fed on a diet free from vitamin A gave no band in the region of $325\mu\mu$, while the liver oils of rats given in addition either crystalline carotene or substances containing carotene showed selective absorption in this region in every case. The absorption spectra shown by the liver oils of the rats treated with carotene were therefore similar

Fig. 1. - - - - Liver oil of rat no. 10 in chloroform.
 Liver oil of rat no. 22 in chloroform.
 ——— Liver oil of rat no. 16 in ether.
 - · - · Liver oil of rat no. 20 in ether.

ϵ is defined by $\log I_0/I = \epsilon cd$, where c is the concentration of the liver oil in g. per cc., d is the cell thickness in centimetres.

to those shown by cod-liver oils in which, as well as in other animals, vitamin A activity has been found to run parallel with a prominent band in the position stated [Takahashi *et al.*, 1925; Morton and Heilbron, 1928; Drummond and Morton, 1929]. Since it has already been shown [Capper, 1930] that this band is absent from the absorption spectrum of carotene (subsequent experiments have also shown that it is absent from the spectrum of red palm oil) it is obvious that the substance responsible (presumably vitamin A) must be synthesised *in vivo* from the carotene.

This result not only affords support to the view that carotene is converted to vitamin A *in vivo* but in a reciprocal manner serves also to strengthen confidence in the reliability of the $325\mu\mu$ band as an index of the presence of the vitamin. Morton and Heilbron [1928] have shown that this band is always present in sources of vitamin A of animal origin, and that when physiological

activity is destroyed by oxidation the band disappears. From the present work additional evidence may be deduced, since when the vitamin, as characterised by its other attributes, is synthesised in the rat, the band simultaneously makes its appearance.

In addition to the main band at $325\mu\mu$ shallow bands or inflexions were noted in the region of $280\mu\mu$. Absorption in this position has been repeatedly described by various workers in the case of cod-liver oils [Schlutz and Morse, 1925; Schlutz and Ziegler, 1926; Heilbron, Kamm and Morton, 1927; Woodrow, 1928]. It has been suggested that this band may be due either to the presence of ergosterol or to a decomposition product of vitamin A, but nevertheless the actual significance remains obscure. From the present work it may be noted, without comment, that inflexions were observed even in the case of the liver oils of rats killed during acute vitamin A deficiency.

SUMMARY.

1. The band at $325\mu\mu$ generally attributed to vitamin A was absent from the absorption spectra of the liver oils of rats suffering from vitamin A deficiency, but was shown by the liver oils of similar rats which had been subsequently cured by massive doses of carotene.

2. Since the $325\mu\mu$ band has already been shown to be absent from the absorption spectrum of carotene, it is obvious that the substance responsible for the exhibition of this band by the liver oils of carotene-treated rats (presumably vitamin A) had been synthesised *in vivo* from the carotene.

3. Additional evidence in favour both of the conversion of carotene to vitamin A and of the reliability of the $325\mu\mu$ band as an index of the presence of the vitamin is therefore made available.

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